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Title: "Cosmetic and/or dermatological compositions containing polyphenols stabilized by perfluoropolyether phosphates and use of perfluoropolyether phosphates as stabilizing agents for polyphenols"

DESCRIPTION

5 Field of application

The present invention refers, in general, to the cosmetic sector of industry.

The invention concerns, in particular cosmetic or dermatological compositions containing polyphenols stabilized by PFPE phosphates and the use of perfluoropolyether (PFPE) phosphates as stabilizing agents for
10 polyphenols.

State of the Known Art

The polyphenols constitute a very vast family of natural substances of vegetal origin, comprising various sub-families like the flavones, flavanones, flavonoids and isoflavones. The first indication concerning the
15 possible biological nutritional and pharmacological function of polyphenols came from Szednt-Gyorgy, the discoverer of Vitamin C, who observed that polyphenols protect Vitamin C from oxidation.

Therefore it has been the antioxidant function of polyphenols that has been the thread leading to very active research over the last fifty years.
20 Even though there has never been found any proof of a vitamin nature for the polyphenols, and hence no characteristic of essentiality in the prevention of a specific syndrome connected with vitamin insufficiency, the marked antioxidant activity shown by many polyphenols has stimulated studies that have resulted in the expansion of their practical
25 application in fields ranging from that of alimentation to those of nutraceuticals and cosmetics.

In fact it is generally accepted today that polyphenols reduce the risk of chronic-degenerative diseases (including cancer) through a series of molecular mechanisms directly or indirectly connected with antioxidant
30 activity.

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In this context one can also include tocopherol (Vitamin E) that, despite its specific vitamin activity on the model of fetal reabsorption, must be considered as a polyphenol of vegetal origin, its assumption reducing the risk of specific chronic degenerative diseases through a mechanism that is, though not exclusively, the antioxidant type.

In general terms antioxidants act in a physio-pathological condition called "oxidative stress", where the balance between the oxidants and the molecules that oppose oxidation favors the former. Oxidants are, in general, free radicals produced by oxygen metabolism under varying conditions, from irradiation with ionizing or UV radiation, to exposure to toxic agents or polluting conditions and inflammation.

Antioxidants interrupt the chain of harmful events at various levels: intercepting the primary oxidant species and those species propagating reaction chains, and governing cell response to the initial lesion. In fact, biological damage from oxidative stress is mostly manifested as a "reaction to the lesion", with a reprogramming of the genic expression of the interested cells. Modified biological behavior, including inflammation and apoptosis, is part of the phenomenology of the damage that can be demonstrated clinically. Skin is a tissue particularly exposed to oxidative stress. In addition to specific inflammatory phenomena, similar to those in other organs, skin is exposed to environmental stimuli that have, generally speaking, the characteristics of oxidants. In fact, UV radiation and a great many environmental pollutants, particularly if subjected to redox transitions like, for example, metals, produce oxidation through an oxyradical mechanism.

The biological reaction to continuous radiation and/or environmental oxidative damage is the first cause of skin aging where the continuously irritating stimulus produces a cell reaction that leads to, in addition to inflammation, protease activation that, when the mechanisms of continuous damage repair are not functionally optimal, finally causes atrophy of the connective tissue matrix and wrinkle formation.

The biological antioxidant defence mechanisms of the skin comprise enzymatic and intracellular chemical systems to which a particular vitamin E cycle is added. In fact the vitamin is secreted with the sebum,

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and this secretion is stimulated by irritative oxidant stimuli like, for example, UV radiation. From the superficial sebaceous layer the vitamin E is therefore reabsorbed through the corneous layer to different deeper depths, so it can be deduced that there is a function also at an extra-cellular level. Experimental evidence therefore suggests that also physiologically there is a mechanism of protection based on "topic" antioxidant use.

The topic use of antioxidants in cosmetology and in slowing down skin aging is supported by the molecular mechanism of the damage and of the antioxidants.

Topically administered antioxidants can be expected to:

- 1) block the initiating oxidant species;
 - 2) block the progression of the oxidative reaction chain;
 - 3) "control" the inflammatory reaction;
 - 15 4) modulate the genic reprogramming responsible for possibly damaging response;
 - 5) inhibit the proteases that degrade the connective tissue matrix;
 - 6) promote cell and tissue repair mechanisms including revascularization.
- 20 To these chemical or biochemical effects there are associated, for different antioxidants, in particular topical formulations, physical effects like the quenching of electronically excited species, the absorption of UV radiation and the prevention of transdermal water loss.

25 Polyphenols are able to exert the beneficial effects illustrated above but
they have the inconvenience of marked instability in the dermatological and/or cosmetic compositions containing them.

In the prior art different methods are proposed to stabilize polyphenols. For example, patent application EP 995 432 suggests the use of surfactants constituted by mono-, oligo- and poly-ethers, esters and ether-esters with alkyl or alkenyl or hydroxyacyl groups with 10-30 carbon

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atoms to stabilize the flavones, flavanones and/or flavonoids against photochemical and/or oxidative degradation.

For the same aim other documents describe the use of an amphiphilic phyllosilicate (EP 1 200 042), of nitrylotriacetic acid (EP 995 422), of 2-
 5 tert-butylhydroquinone (EP 997 133), of saturated fatty acid mono- and/or diglyceride esters partly neutralized with citric acid (EP 1 000 603), of an alkylglucoside tensioactive (EP 998 898) and of butylhydroxytoluol (EP 998 899).

Summary of the invention

- 10 The problem at the base of the present invention is to provide a new stabilizing agent for the polyphenols, allowing the achievement of cosmetic or dermatological formulations for topical use, in which the polyphenols are stable and active throughout the whole conservation period normally required for such products that is at least 36 months.
- 15 Such a problem has been solved using as the said polyphenol stabilizing agent at least one perfluoropolyether phosphate (PFPE phosphate), in particular a perfluoropolyether phosphate of formula (I):



wherein

- 20 $x = 1$ or 2 ;

R_1 and R_2 are independently selected from H and CH_3 ;

n is an integer between 1 and 50, preferably 1-6;

- R_f is a perfluoropolyether chain with a number average molecular weight between 400 and 1800, preferably 500-1300, comprising repeating units
 25 chosen from among the following:

a) $-(C_3F_6O)-$

b) $-(CF_2CF_2O)-$

c) $-(CFL_0O)-$, wherein $L_0 = -F, -CF_3$;

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d) $-\text{CF}_2(\text{CF}_2)_y\text{CF}_2\text{O}-$, wherein $y = 1$ or 2 ;

e) $-\text{CH}_2\text{CF}_2\text{CF}_2\text{O}-$,

and wherein, when $x = 1$, an end group is a perfluoroalkyl selected from CF_3O , $\text{C}_2\text{F}_5\text{O}$, $\text{C}_3\text{F}_7\text{O}$.

5 Particularly preferred are the perfluoropolyether phosphates of formula (I) in which R_f has one of the following structures:

1) $-(\text{CF}_2\text{O})_a-(\text{CF}_2\text{CF}_2\text{O})_b-$

wherein b/a is between 0.3 and 10 and a is an integer different from 0;

2) $-(\text{CF}_2-(\text{CF}_2)_y-\text{CF}_2\text{O})_{b'}-$

10 wherein $y = 1$ or 2 ;

3) $-(\text{C}_3\text{F}_6\text{O})_r-(\text{C}_2\text{F}_4\text{O})_b-(\text{CFL}_0\text{O})_t-$,

wherein $r/b = 0.5-2.0$, $(r+b)/t = 10-30$, b and t are integers different from 0;

4) $-(\text{OC}_3\text{F}_6)_r-(\text{CFL}_0\text{O})_t-\text{OCF}_2-\text{R}'_f-\text{CF}_2\text{O}-(\text{C}_3\text{F}_6\text{O})_r-(\text{CFL}_0\text{O})_t-$

15 5) $-(\text{CF}_2\text{CF}_2\text{CH}_2\text{O})_{q'}-\text{R}'_f-\text{O}-(\text{CH}_2\text{CF}_2\text{CF}_2\text{O})_{q'}-$

where R'_f is a fluoroalkylene group with 1-4 carbon atoms;

L_0 is selected between F and CF_3 ;

6) $-(\text{C}_3\text{F}_6\text{O})_r-\text{OCF}_2-\text{R}'_f-\text{CF}_2\text{O}-(\text{C}_3\text{F}_6\text{O})_r-$

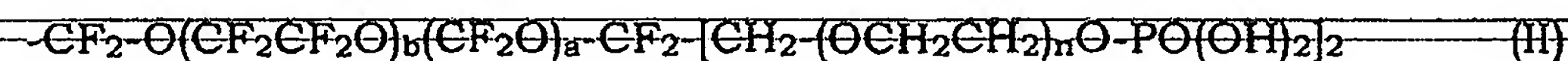
wherein in the above formulas:

20 $-(\text{C}_3\text{F}_6\text{O})-$ represents units of formula:

$-(\text{CF}(\text{CF}_3)\text{CF}_2\text{O})-$ and/or $-(\text{CF}_2-\text{CF}(\text{CF}_3)\text{O})-$

a , b , b' , q' , r , t are integers whose sum is such that R_f shows values of number average molecular weight M_n that fall between about 400 and 1800, preferably between 500 and 1300.

25 Particularly advantageous to the goals of the present invention is the use of perfluoropolyether diphosphates of the general formula (II):



wherein $n = 1$ or 2 , $b/a = 0.5-3.0$ and a , b and r have the above reported meanings.

30 The above perfluoropolyether phosphates and diphosphates are known from patent applications EP 1 074 243 and EP 1 145 722, that respectively describe their use as cosmetic composition ingredients of high

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water- and oil-repellency and as preservatives in formulas for topical use, without mentioning any stabilizing effect with regard to polyphenols, to vitamin E or to other compounds with antioxidant activity.

Using the said perfluoropolyether diphosphates as stabilizing agents, there have been provided, according to the present invention, cosmetic and/or dermatological compositions for topical use comprising as the active substance polyphenols associated with a suitable carrier, characterized by containing, as the stabilizing agent, an effective amount of a perfluoropolyether phosphate, in particular a perfluoropolyether diphosphate according to the formula (II).

Preferably, such cosmetic and/or dermatological compositions contain a perfluoropolyether diphosphate amount of between 0.1 and 5.0% in weight of total composition weight and conveniently from 0.2 to 1.0%.

The polyphenol content is preferably between 0.1% and 5% by weight of the total composition weight and conveniently from 0.2 to 2%.

The Applicant has furthermore ascertained that perfluoropolyether diphosphates stabilize, very efficiently, another natural antioxidant, vitamin E, also in its non-esterified form.

Therefore the present invention concerns, furthermore, cosmetic and/or dermatological compositions for topical use containing, as active substances, one or more polyphenols and vitamin E, together with a suitable carrier, characterized by containing as stabilizing agent an effective amount of a perfluoropolyether diphosphate according to the formula (II).

Vitamin E can be used as d- α -tocopherol or as a mixture of the two enantiomers d and l of the α -tocopherol or as a mixture of other tocopherols (β , γ , ϵ , ζ , η) of vegetal origin or as tocotrienol. The different forms of vitamin E can be natural or synthetic in origin.

The amount of perfluoropolyether diphosphate contained in such compositions is that indicated above.

Different vegetal polyphenols and Vitamin E can sustain, with different efficiency, protection mechanisms against free radicals and oxidative

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stress, so for this reason the association of one or more polyphenols with vitamin E is particularly effective.

On the basis of the specific reactivity of the different polyphenols it is foreseen that particularly efficient skin protection should result from a mixture of a) Vitamin E in the free form; b) one or more polyphenols, like, for example, a standard extract of grape seed rich in polymeric procyanidins and an extract of green tea rich in epigallocatechin gallate.

A topical preparation using these components is very difficult to obtain for two reasons: the different lipophilicity/hydrophilicity and solubility of the components and the susceptibility to oxidation, this last an obvious and inevitable consequence of the high antioxidant activity.

The redox chemistry of phenolic antioxidants foresees that autoxidation begins with the extraction of an atom of phenolic hydrogen or, far more easily, with a one electron transition permitted by the acid dissociation of the phenol. This reaction is furthermore favored by electron acceptors like transition metals and the presence of water at the reaction site.

Therefore the stabilization of the polyphenols and vitamin E for a cosmetological preparation requires an environment in which the phenol groups are not exposed to water, and an acidity that prevents their dissociation.

The PFPE diphosphate is an acid ester as it is obtained by monosubstitution of orthophosphoric acid. The acidity of the most active hydrogen acid is even higher than that of the hydrogen with the greatest protonic characteristics of phosphoric acid, as is demonstrated by the comparison between the corresponding dissociation constant values (PFPE phosphate $\text{PK}_a = 1.84$; orthophosphoric acid $\text{pK}_a = 2.15$).

~~PFPE diphosphate is very soluble in alcohols and glycols, also in a ratio of~~
1:1. Unexpectedly the addition of water to a concentrated solution of PFPE phosphate and alcohol or glycol leads to the formation of limpid solutions with an acidic pH; in concentrations between 0.5 and 5% they have a pH of 2.0-3.5. As demonstrated by numerous dermatological patch tests, these solutions or the emulsions that contain them have, despite the acidity, been shown to be non-irritating at the cutaneous level and even

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capable of reducing the cutaneous irritation of other substances or other acids like the alpha-hydroxyacids.

Therefore acidifying the emulsion with PFPE phosphates results in a preparation, not irritating at the cutaneous level, in which the polyphenol
5 substances present for example in the extract of green tea or grape seeds are, like tocopherol, stabilized.

The optimum stability demonstrated, also under heating, by the emulsion in question leads to the supposition of a stabilizing mechanism that is not simply due only to the acid environment, but also to an active role of the
10 fluorinated substance, which is able to protect the vegetal extracts and tocopherol, due to the perfluoropolyether chain having a screening effect with regard to the phenolic molecules.

Furthermore the compositions according to the present invention can optionally contain other compounds endowed with antioxidant or else
15 vitaminic activity and particularly susceptible to oxidation, like for example vitamin A, carotenes, carotenoids, lutein, lycopene and xanthophylls.

It is to be noted that even free ascorbic acid is surprisingly stabilized by the perfluoropolyether phosphates, even though it is notoriously unstable
20 in the usual cosmetic and dermatological formulations.

The stabilization of ascorbic acid is achieved with the use of perfluoropolyether phosphates according to the invention, in particular with those according to formula (II), in the percentage amounts reported above in relation to polyphenol stabilization.

25 Detailed description of a preferred embodiment

The present invention will be further described making reference to several examples of formulations according to the present invention, supplied here as illustrative and not limiting, in which the different ingredients are indicated with the respective name INCI and in percentages of weight on
30 total weight.

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EXAMPLE 1

	Steareth-2	4
	Steareth-21	4
	Cetearyl alcohol	4
5	Glyceryl stearate	3
	Octyldodecanol	3
	Dimethicone	0.5
	Tocopherol	5
	Glycerin	8
10	Pentylene Glycol	7
	Disodium EDTA	0.05
	Polyperfluoroethoxymethoxy-difluoroethyl PEG phosphate ¹	0.5
	Camelia sinensis ²	0.5
	Vitis vinifera ³	0.5
15	Aqua	q.b. a 100

1) Use was made of the commercial product Fomblin HC/P2-1000® of the company Solvay Solexis.

2) Use was made of the Indena company product Greenselect®, a stabilized extract of green tea.

20 ~~3) Use was made of the Indena company product Leucoselect®, a stabilized extract of grape seeds.~~

A cream was prepared by putting into an emulsifier first the water and the water soluble ingredients, except for Greenselect® and Leucoselect® and then all the oily and liposoluble ingredients, except tocopherol (free
25 vitamin E) and heat was applied until the fats had melted completely.

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Successively vitamin E was introduced and the vacuum pump activated to obtain a pressure of 40 cmHg. At this point the turbine and the mixer of the emulsifier were put into action at maximum speed for 10-15 minutes, after which cooling to room temperature took place; with the turbine
5 turned off the polyphenols (Greenselect® and Leucoselect®) were added, and mixed in for several minutes.

The thus obtained O/W cream was beige-orange in color, with pH 3.7, a viscosity (10 rpm) of 32,000 mPs s and stable at a centrifugation of 6000 rpm for 30 minutes.

10 The cream was packed in plastics containers of 50 ml and subjected to a test to confirm the stability. A series of samples of the cream contained in the said small plastic jars was kept in an oven at 45°C for twelve weeks, after which the viscosity of the cream was checked and found unaltered with respect to the starting value.

15 In addition there was the verification of the amount of polyphenols and vitamin E present in the cream at the end of the twelve weeks at 45°C, by means of an HPLC method, using a Macherey-Nagel column, Nucleosil 100-5 C18, packed with 5 µm particles (150 x 4.6 mm internal diameter), and a pre-column system Nucleosil 100-5 C18, CC 8/4.

20 The determination of free tocopherol, carried out using methanol as the eluent in isocratic conditions, with a flow rate of 1 ml/min and λ 290 nm, gave a value of 5.2%.

The determination of the polyphenol concentration in the cream was carried out using a mobile phase in binary gradient as eluent, composed
25 of: phase A = 0.3% formic acid in water and phase B = methanol; flow rate 1 ml/min; λ = 278 nm. In such conditions the Greenselect® has a retention time R_t = 22.84 min and the Leucoselect® R_t = 6.53 min.

The above determination supplied a value of 0.50% for Greenselect® and 0.49% for Leucoselect®.

30 As can be easily ascertained from the above reported data, the concentration of the three active ingredients (vitamin E and the two polyphenols) remained substantially unaltered after a twelve week period

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at 45°C. This confirms that the presence of the perfluoropolyether diphosphate in the cream stabilized both the vitamin E and the polyphenols with regard to oxidative degradation.

EXAMPLE 2

5	Steareth-2	4
	Steareth-21	2
	Cetearyl alcohol	5
	Glyceryl stearate	6
	Octyldodecanol	6
10	Dimethicone	0.5
	Glycerin	6
	Pentylene Glycol	7
	Disodium EDTA	0.06
	Polyperfluoroethoxymethoxy-difluoroethyl PEG phosphate ¹	0.8
15	Camelia sinensis ²	1.0
	Vitis vinifera ³	1.0
	Aqua	q.b. a 100

For the references 1, 2 and 3 see what is reported in example 1.

An O/W cream was prepared starting with the ingredients listed above
20 and proceeding in a way analogous to example 1.

The thus obtained O/W cream was beige-orange in color, had a pH of 3.8, a viscosity (10 rpm) of 31,000 mPs s and was stable to centrifugation at 6000 rev/min for 30 minutes.

The cream, subjected to the same stability verification test described in
25 example 1, was shown to be perfectly stable and to conserve, substantially

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unaltered, the polyphenol content.

EXAMPLE 3

	Steareth-2	3
	Steareth-21	4
5	Cetearyl alcohol	4
	Glyceryl stearate	3
	Octyldodecanol	3
	Dimethicone	0.5
	Tocopherol	4
10	Glycerin	8
	Pentylene Glycol	7
	Ascorbic acid	2
	Disodium EDTA	0.08
	Polyperfluoroethoxymethoxy-difluoroethyl PEG phosphate ¹	1.0
15	Camelia sinensis ²	0.8
	Vitis vinifera ³	0.8
	Aqua	q.b. a 100

For the references 1, 2 and 3 see what is reported in example 1.

20 An O/W cream was prepared starting with the ingredients listed above and proceeding in a way analogous to example 1.

The thus obtained O/W cream was beige-orange in color, had a pH of 3.6, a viscosity (10 rpm) of 32,000 mPs s and was stable to centrifugation at 6000 rev/min for 30 minutes.

The cream, subjected to the same stability verification test described in

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example 1, was shown to be perfectly stable and to conserve, substantially unaltered, the content of the polyphenols and vitamin E.

The stability of the ascorbic acid contained in the cream was determined as follows.

- 5 A series of samples of the cream contained in the said small plastic jars was kept for a period of 4 months at room temperature and then in an oven at 45°C for one month, after which the ascorbic acid content of the cream was checked.

- 10 In order to determine the ascorbic acid content, 100 mg of cream were diluted in 100 ml of 0.1 M phosphate buffer at pH 6, and the thus obtained suspension was magnetically stirred in the dark for 20 minutes. Thereafter, 30 ml of the suspension were transferred to a 50 ml centrifuge tube and 15 ml of chloroform were added. The tube was shaken for 10 minutes and then centrifuged at 6000 rpm for 5 minutes. The upper
15 aqueous phase was directly analyzed with a UV Vis Jasco V-350 spectrophotometer, using phosphate buffer as a blank and the ascorbic acid peak at 266.5 nm was measured.

- The ascorbic acid concentration was determined by means of a calibration curve made from standard solutions containing 1.0, 1.5 and 2.0 mg
20 ascorbic acid/100 ml phosphate buffer.

- The absorbance value obtained for a solution of 2 mg of ascorbic acid in 100 ml phosphate buffer was 1.619 and the value obtained for the sample prepared as described above was 1.492. Based on the calibration curve, the latter value corresponded to an ascorbic acid concentration of 1.844
25 mg/100 ml.

This means that only 7.8% of ascorbic acid was lost by the end of the storage period as specified above.
